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Effects of acid rain components on soil microarthropods: A field manipulation

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Summary. The effects of a range of nitrogenous and sulphuric pollutants, applied as simulated rainfall, on microarthropod assemblages in a *Picea abies* stand in Co. Kilkenny, Ireland, were investigated. The treatments were: nitric acid, sulphuric acid, ammonium sulphate, ammonium nitrate and ammonium chloride. Urea and streptomycin sulphate (a bacteriocide) were applied to extend the range of contrasts. Microarthropods were sampled one and two years after the initiation of spraying. Effects were observed on 29 species of the 140 species enumerated, but the alterations in the abundance of these affected species were rarely consistent on both sampling dates. When the abundances of individual species were aggregated into higher level taxa or into trophic/functional groups the impact of the pollutants was more discernible. Collembola, Crypostigmata, Asigmata and Prostigmata were affected by two or more treatments. Most changes in abundance relative to control plots were increases. Microphytophages (microbial feeders) were the most widely affected functional group. Abundance increased in all plots other than ammonium chloride and streptomycin. This is consistent with the notion that the effects on microarthropod abundance are mediated by the more sensitive microbial community. Ordination and Monte Carlo Permutation tests revealed that the assemblages of microarthropods in the organic layer of nitric acid sprayed plots differed from those from other nitrogen receiving plots and that the assemblages in the nitric acid treated plots differed from those in the sulphuric acid plots. In the mineral soil, after one year, each plot contained a unique assemblage when they are contrasted with the control plots, but this result was not repeated in the second year. Results when predators are considered mirror the patterns noted for the microphytopages. Diversity indices and the use of bait strips, to monitor feeding activity, confirm that the strongest differences emerged from the nitric and sulphuric acid plots. The results are found to be consistent with the hypothesis that responses to the components of "acid rain" will be determined as much by their nutrient content and chemical form of the application as by their acidifying potential.

Key words: Acari, Collembola, microarthropods, acid rain, perturbation, community structure

Introduction

The high densities maintained by many animal groups in the soil implicate them in important regulatory roles in nutrient cycling. Budgetary studies show a faunal contribution of approximately 30% to nitrogen mineralization in many systems (Rosswall & Paustian

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1984; Hunt et al. 1987). Changes in faunal assemblage structure may, by implication, have consequences for the maintenance of soil fertility and plant growth (Wright & Coleman 1988; Setälä & Huhta 1991). In a coniferous forest, which is the focus of this study, the importance of the acarine and collembolan fauna is accentuated by the relative scarcity of annelids.

Increased anthropogenic emissions of sulphur and nitrogen and their transformation in the atmosphere into strong acids which can be deposited in a wet or dry phase has received much attention (Likens 1989). Concerns about the effects of such deposition on forests have centred on their effects on soil properties (Eriksson et al. 1992; Abrahamsen & Struanes 1986), nutrient cycling (Johnston et al. 1985), tree growth (Tamm & Cowling 1977) or their impact on microbial activity and assemblage composition (Hovland 1981; Arnebrant et al. 1990; Wookey et al. 1991; Rühling & Tyler 1991). In addition to the acidifying effects of this pollution the impact of the increased nutrient depositions has been discussed (Nihlgård 1985; Aber 1992).

In contrast, the investigation of the effects of such acidification and nutrient pollution on soil microarthropods has been minimal. Investigations of acid precipitation have focused primarily on the effects of sulphuric acid, reflecting the concerns of the Nordic countries where these studies have been carried out (Bååth et al. 1980; Hågvar & Abrahamsen 1980; Hågvar & Amundsen 1981; Hågvar & Kjøndal 1981; Hågvar 1984). The impact of nitrogen compounds on soil fauna has been studied predominantly with a view to determining the effects of silvicultural practice (Huhta et al. 1983; Huhta et al. 1986; Vilkamaa & Huhta 1986; Koskenniemi & Huhta 1986; Lohm et al. 1977). Consequently, in research, nitrogenous compounds have been applied to the forest floor in single, or less frequently in two or three, applications and the effects on the fauna are followed for a number of years subsequent to the application (e.g. Marshall 1974; Behan et al. 1978; Fratello et al. 1989; Lohm et al. 1977). The effect of such large single doses is often a 'shock effect', a consequence of altered osmotic potential (Heugens & van Daele 1984) and some fertilizers are known to be toxic to microarthropods when applied in high concentrations (Huhta et al. 1986).

In this study we investigated the impact on microarthropods (Acarina and Collembola) of a range of nitrogenous and sulphuric pollutants applied in a simulated rainfall. Three of these: nitric acid, sulphuric acid and ammonium sulphate are known components of acid precipitation and have been shown to acidify soil. Three others: ammonium nitrate, ammonium chloride and urea are nitrogen compounds which decrease the soil pH to a lesser extent or may even raise it (at least temporarily). The hypothesis being explored is that the effects of different components of polluted rain will be determined as much by their nutrient content and by the chemical form in which they are deposited as by their acidifying power. Streptomycin and glucose were applied to soil to investigate the effects of altering the microbial assemblage on the soil fauna.

The rationale of the experiment, therefore, is in contrast to most research on acid rain or on nitrogen impacts on microarthropods where the applications are commonly in single large applications with amounts of up to 460 kg N/ha being used. The methods employed here are intended to explore deposition from rain rather than the application of fertilizer and the wide range of treatments employed permit a greater range of contrasts to be made. An approach similar to this was employed by Craft & Webb (1984) where they contrasted the effects of acid and neutral sulphate salts on forest floor arthropod trophic groups.

Materials and Methods

Site Description

This field investigation was carried out in a unthinned *Picea abies* stand 10 km west-south-west of Kilkenny on the south facing slope of the Slieveardagh Hills (Latitude 52°27.05 N, longitude 07°23.49 W). The stand was planted in 1968 on a gleic brown earth at a density of 2500 stems ha^{-1}.

The tree height at the time the experiment was performed was approximately 12 m (growth class 14). The canopy cover approximated 90% and there was no ground vegetation. The annual precipitation is 826 mm and the annual temperature 9.4 °C (from Kilkenny weather station 1957 – 1980). The annual throughfall input at the site contained 2.65 kg NO_3-N, 5.54 NH_4-N, 8.2 kg SO_4-S and .01 kg H^+ (McCarthy pers. comm.).

Experimental protocol

Seven treatments were applied, nitric acid, sulphuric acid, ammonium sulphate, ammonium nitrate, ammonium chloride, urea and streptomycin-sulphate plus glucose. As a control, distilled water was applied. Treatments were applied in a randomized block design with three blocks. The nitrogen containing compounds were applied in amounts corresponding to 150 kg N $ha^{-1} \cdot a^{-1}$ and the amount of sulphuric acid was applied to give an amount of sulphate equal to that in ammonium sulphate (i.e. 171.4 kg S $ha^{-1} \cdot a^{-1}$). At each application streptomycin sulphate and glucose were applied in the ratio of 72 g streptomycin to 24 g glucose. The acids and salts were dissolved in 1 l of water and sprayed in a fine mist on plots measuring 6 m^2. The pH of the sprayed compounds is given in Table 1. Treatments were applied in 24 monthly increments starting on 21 March 1989 and continuing until 7 May 1991.

Sampling regime

Faunal sampling was carried out on two occasions: 30 March 1990 and 7 May 1991, approximately one and two years, respectively, after the initiation of spraying. On each occasion three samples were taken from each plot using a steel corer. The samples were 25 cm^2 in area and were divided into the organic layer and the top 6 cm of the mineral soil layer. Microarthropods were extracted, using a Macfadyen high gradient extractor over a period of one week, into plastic cups containing picric acid. 95% I.M.S. was added to preserve the organisms. Where possible all Acarina and Collembola were identified to species level.

Bait strips

Bait strips (von Torne 1990) were inserted in the soil to estimate animal activity towards the end of the experiment (25 April 1991). These strips, constructed from PVC plates, measured 6 × 150 × 0.5 mm are perforated 16 times at 5 mm intervals. Bait (comprised of a substrate of 83% sieved chernozem soil with 5% gypsum as a binding agent, 5% powdered bran flakes, 4.7% powdered milk, 2% peptone and 0.3% mannitose) was compressed into each perforation and the strips were placed in groups of 16 vertically into the soil such that the upper perforations were flush with the surface of the organic horizon. The strips remained in the field for 72 hours. The strips were viewed under a compound microscope and both the number of perforations from which bait has been removed by faunal feeding and the depth profile of the removals were assessed.

Table 1. pH, nitrogen and sulphur content of the sprays applied and pH of the organic layer at the end of the experiment

	pH of application	Amount of N or S applied (kg $ha^{-1} \cdot a^{-1}$)	pH or organic layer at end of experiment
Nitric Acid (HNO_3)	.7	150 N	4.5
Sulphuric Acid (H_2SO_4)	1.0	171.4 S	4.7
Ammonium Nitrate (NH_4NO_3)	5.2	150 N	4.8
Urea ($CO(NH_2)_2$)	7.4	150 N	5.0
Ammonium Sulphate ($(NH_4)_2SO_4$)	5.4	150 N and 171.4 S	5.3
Ammonium Chloride (NH_4Cl)	5.1	150 N	5.3
Control (H_2O)	5.9	0	5.3
Streptomycin & glucose	5.9	–	6.2

Organic layer pH

The pH of the organic layer (in H_2O) was measured with a WTH pH 522 pH probe at the end of the experiment.

Data analysis

Differences in the abundances of individual species and groups were investigated using analysis of variance of $\log_{10}(n + 1)$ transformed data. The data were analyzed as a randomized block design. Since the three replicate faunal samples per plot taken on each sampling occasions were not independent estimates of faunal abundance (and were pseudoreplicates, sensu Hurlbert 1984), the treatment by block interaction term was used as the error term in calculating F values for testing the hypothesis that changes in taxa were associated with the applied treatments. The following pre-planned comparisons were investigated using contrast analysis (contrast analysis of variance permits a deeper evaluation of multiple treatment experiments (Rosenthal & Rosnow 1985)):

- Differences between each treatments and the control
- Differences between all nitrogen-containing treatments
- Differences between all ammonium releasing treatments
- Differences between sulphur-containing treatments
- Differences between the two strong acids (nitric and sulphuric acid)
- Differences between the acids and other treatments
- Differences between sulphur and nitrogen treatment

Since the dry weights of the organic layer and soil samples from which the animals had been extracted were found to vary, changes in the proportions of the extracted taxa from the samples were tested for differences. The results of this analysis did not differ from those obtained using absolute abundance and are therefore not presented.
Species richness (R), and the log series index (Fisher's α), were calculated to assess the impact of the treatments upon faunal diversity. The log series index is chosen as a measure of diversity since it has good discriminant ability but is relatively insensitive to sample size. Its use is recommended whether or not the log series model is the most appropriate for describing the underlying species abundance distribution (Magurran 1988). The score is given by the equation:

$$\alpha = \frac{N(1 - x)}{x}$$

where x is the iterative solution of $S/N = (1 - x)/x[-\ln(1 - x)]$.
Species richness (the number of species per sample) is appealing because of its intuitive simplicity.
The values obtained from these scores, when $\log_e$ transformed, were found to satisfy the conditions for analysis of variance. Differences between each treatment and the control were tested and differences amongst the ammonium-containing, nitrogen-containing and acid treatments were tested.
Responses of the microarthropod assemblages to the treatments were analyzed using canonical correspondence analysis CCA. CCA is a multivariate direct gradient technique whereby a set a species is related to a set of environmental variables. The technique identifies an environmental basis for community ordination by detecting the patterns of variation in assemblage composition that can be best explained by the environmental variables. It thus allows a quick appraisal of how assemblage varies with the environment (ter Braak 1986). This analysis is accompanied by Monte Carlo permutation tests, performed using the CANOCO program, which allow tests of significance of the axes in the ordination.

Results

Organic layer pH

The pH of the litter sprayed with nitric acid, sulphuric acid, ammonium sulphate and ammonium chloride was lower than from the control plots at the end of the experiment. Urea and ammonium nitrate were similar in pH to the control and streptomycin and glucose treated plots were higher (Table 1).

General comments on the fauna

There were 49,309 microarthropods recovered from the organic layer and soil samples collected. The most abundant group were the Crytostigmata which were represented by 42 species. In addition, there were 44 species of Mesostigmata, 21 of Prostigmata and 5 of Asigmata. Twenty eight species of Collembola were recovered. One year into the experiment the Collembola dominated the mineral soil fauna and Cryptostigmata dominated in the organic layer. The same situation obtained after two years although, in that year, there were greater numbers of both groups in each horizon (Collembola increased in number by 15% and Cryptostigmata increased by 21%). The total number of Prostigmata was lower by 13% after two years compared with the first year. Lower numbers of Astigmata (25% down) and Mesostigmata (11% down) were also recovered in the second year. There was an overall 22.7% increase in microarthropod numbers after two years.

Four functional groups were identified among the microarthropods occurring in this system, namely, microphytophages, macrophytophages, panphytophages and predators. Species were assigned to these trophic groups on the basis of an extensive literature review of microarthropod trophic activity (O'Connell, unpublished manuscript). Microphytophages feed on bacteria, fungi or algae. Macrophytophages feed on litter, and panphytophagous microarthropods are generalist in their feeding strategy. Predatory microarthropods feed upon a wide range of fauna in the soil.

Microphytophages were represented by more than 80 taxa and comprised over 90% of all animals in both years. The dominance of this functional group was comparable in both organic layer and soil. Predators with over 40 taxa were the next most abundant group and comprised 7.1% of animals after one year and 6.7% after two years. They were more prevalent in the soil than in the organic layer. After one year, 54% of predators were soil-dwelling and after two years their prominence in the soil increased marginally to 58%. Macrophytophages were represented by 9 taxa and constituted 1% of fauna after one year and 1.7% after two years. Consistent with their functional role, in excess of 90% of macrophytophages occurred in organic layer samples in both years. Panphytophages were least diverse (9 taxa) and constituted less than 1% of microarthropods in both years. After one year a majority (84%) were found in the organic layer whereas after two years approximately equal numbers were found in organic layer and soil.

Treatment effects

Supra-specific taxonomic groups. Mean abundances of supra-specific taxonomic groups and trophic/functional groups are shown in Tables 2 and 3. Except when otherwise stated treatment effects discussed are significant at $p \leq .05$. On the first sampling occasion no effect of treatments on the total abundance of Collembola was detected. Differences emerged between plots treated with the strong acids on the second sampling occasion. A lower mean abundance was found in nitric acid plots than in sulphuric acid plots (i.e. 12,400 m^{-2} in nitric acid plots compared with 41,200 m^{-2} in sulphuric acid plots). This represented a 48% decrease in mean abundance in nitric acid plots compared with the control plots (which had a total mean abundance of 19,000 m^{-2}) and an increase of 60% over control plots after treatment with sulphuric acid (although differences from control plots, in the case of these latter two comparisons were not statistically significant).

After one year, abundances of Cryptostigmata were more than double control levels in the organic portion of ammonium sulphate treated (i.e., a mean of 16,100 m^{-2} in controls and 52,200 m^{-2} in ammonium sulphate plots). Although numbers remained relatively high in the second year, the differences were not significant. Abundance was also high in the organic portion of ammonium nitrate treated plots compared to the controls after one and two years. The differences were significant in year two only, when the mean abundance was 63,000 m^{-2} in the treated plots compared to 26,700 m^{-2} in control plots.

Table 2. Toral abundance in 1000's (means per m^2) of supra-specific taxonomic and trophic/functional groupings associated with experimental plots in organic (O) and mineral (M) layer after one year

Group	Treatment															
	Nitric Acid		Ammonium Sulphate		Sulphuric Acid		Ammonium Nitrate		Urea		Control		Ammonium Chloride		Streptomycin	
	O	M	O	M	O	M	O	M	O	M	O	M	O	M	O	M
Taxonomic Group																
Acari																
Cryptostigmata	33.7	9.0	52.2	17.9	27.7	9.2	24.1	20.3	27.5	27.7	16.1	7.4	23.4	3.8	17.2	9.0
Mesostigmata	4.4	1.9	5.0	2.5	5.1	1.8	6.7	3.3	5.7	2.2	6.4	2.4	4.4	1.4	6.7	2.5
Prostigmata	9.7	21.3	13.5	16.3	16.8	10.8	10.9	13.9	10.6	24.7	8.3	11.6	17.9	14.3	8.1	5.1
Astigmata	6.0	2.9	4.6	.59	4.4	.54	5.3	.81	5.8	.68	5.8	.18	5.6	.04	5.0	.72
Collembola																
Total Collembola	14.5	45.3	17.9	29.9	14.6	23.5	18.4	33.7	15.2	29.0	19.0	17.9	15.4	26.1	10.5	16.9
Functional Group																
Microphytophages	63.0	74.3	87.4	62.5	63.6	41.5	58.3	67.8	58.4	79.5	48.4	34.5	62.2	38.9	39.9	30.1
Macrophytophages	1.5	.04	.77	0	1.2	0	1.5	.13	1.7	0	1.2	0	.86	0	1.3	.23
Predators	3.02	6.2	3.4	4.6	3.2	4.3	4.8	3.9	4.01	4.6	5.4	4.8	3.0	6.7	5.6	3.1
Panphytophagues	.72	.05	1.6	.22	.63	.13	.9	.23	.72	.36	.68	.09	.81	.13	.72	.45

Table 3. Toral abundance in 1000's (means per m^2) of supra-specific taxonomic and trophic/functional groupings associated with experimental plots in organic (O) and mineral (M) layer after two year

Group	Treatment															
	Nitric Acid		Ammonium Sulphate		Sulphuric Acid		Ammonium Nitrate		Urea		Control		Ammonium Chloride		Streptomycin	
	O	M	O	M	O	M	O	M	O	M	O	M	O	M	O	M
Taxonomic Group																
Acari																
Cryptostigmata	58.2	12.1	59.2	23.1	62.1	24.8	63.0	15.2	49.0	8.3	26.7	12.4	33.6	12.0	29.7	15.8
Mesostigmata	33.8	1.8	5.1	1.4	2.5	1.0	5.6	1.4	6.7	1.3	6.7	.59	3.8	1.2	6.7	1.1
Prostigmata	11.5	4.8	8.5	12.2	14.0	16.8	18.4	10.0	22.0	9.7	10.1	9.0	11.9	7.0	6.3	3.02
Astigmata	.59	.27	5.23	1.3	.77	1.0	2.8	.9	2.9	.32	1.9	.3	5.3	2.2	2.9	.59
Collembola																
Total Collembola	12.4	35.5	23.2	51.0	41.2	28.1	30.3	40.4	23.4	24.7	25.7	32.3	21.7	21.2	29.2	26.2
Functional Group																
Microphytophages	82.4	50.8	94.2	80.2	113.4	59.7	113.0	60.5	94.6	37.4	62.9	47.5	68.8	38.4	67.6	43.8
Macrophytophages	.5	.04	1.8	.09	3.3	.27	1.8	.27	3.9	.04	2.1	.36	3.3	.13	2.3	.04
Predators	2.9	2.5	4.9	8.7	3.5	10.9	4.8	6.9	5.1	6.1	5.5	5.7	3.7	4.6	4.5	2.03
Panphytophagues	.31	.5	.41	.05	.31	.86	.5	.27	.54	.63	.63	.95	.45	.68	.45	1.1

In the mineral soil portion of nitric acid treated plots the abundance of Astigmata was sixteen times larger than that of the control plots (2,900 m^{-2} in treated plots compared to 180 m^{-2} in the controls), while the abundances in the organic layer were quite similar. After two years, the astigmatid abundance in the organic layer of the ammonium sulphate (5,230 m^{-2}), urea (2,900 m^{-2}), and ammonium chloride (5,300 m^{-2}) was elevated compared with levels in control plots (1,900 m^{-2}). The abundance of astigmatids in acid treated plots were both below 1,000 m^{-2} and significantly lower when contrasted with all other plots. Differences were detected when astigmatid abundance in all nitrogen-treated plots were compared. The low abundance in nitric acid plots was responsible for this as no differences were found between abundances in ammonium-receiving plots. When the abundance in the organic layer and mineral soil were combined the relative enhancement of abundance in ammonium sulphate and ammonium chloride treated plots remained significant, as did the reduction in the acid-treated plots compared with others.
After one year the abundance of Prostigmata in the mineral portion of nitric acid treated plots (21,300 m^{-2}) was twice that of the control plots (11,600 m^{-2}) whereas abundance was approximately halved by applications of streptomycin (5,100 m^{-2}). Differences between abundances in plots treated with the two strong acids were detected as a consequence of this. On the second sampling occasion the mean total abundance in the organic layer of ammonium nitrate (18,400 m^{-2}) and urea (22,000 m^{-2}) treated plots were approximately twice control levels (10,100 m^{-2}). Increases in abundance of prostigmatids in mineral soil portions of ammonium sulphate and sulphuric acid treated plots were also detected. When mineral and organic portions were combined for year two, increased abundance was detected from sulphuric acid, ammonium nitrate and urea treated plots relative to levels in control plots.
The Mesostigmata did not respond significantly to the treatments when abundance in the treated plots were compared with levels in the control.

Functional groups. On the first sampling occasion the abundance of microphytophages in the organic layer of ammonium sulphate treated plots (87,400 m^{-2}) was 80% larger than that of the control (48,400 m^{-2}) and in sulphuric acid plots (63,600 m^{-2}) it was 30% larger. The abundance of this trophic group was more than twice as large in the mineral soil portion of nitric acid (74,300 m^{-2}) and urea (79,500 m^{-2}) treated plots compared with the controls (34,500 m^{-2}). An approximately 80% increase in abundance in organic layer of sulphuric acid (113,400 m^{-2}) and ammonium nitrate treated plots (113,000 m^{-2}) was detected after two years compared to control levels (62,900 m^{-2}). When abundances from the organic layer and mineral soil were combined there was an overall increase in the abundance of microphytophages in ammonium sulphate and ammonium nitrate treated plots. These increases resulted in differences being detected amongst the ammonium-treated plots.
No differences in the abundance of macrophytophages was detected on the first sampling occasion. After two years, the mean adundance of macrophytophages in the organic layer of nitric acid treated plots (i.e. 500 m^{-2}) was significantly lower than that of the control (2,100 m^{-2}). The differences between the acids was significant although when the two strong acids were considered together they differed significantly from other treatments. These differences were also reflected when the mineral and organic portions were combined for year two, although it must be remembered that macrophytophages were very poorly represented in the mineral soil.
The abundance of predators was not affected by treatments in year one. Their abundance in the mineral soil of ammonium sulphate treated plots (i.e. 8,700 m^{-2}) was larger than the control level (5,700 m^{-2}) in the second year. No other impact upon the abundance of predators was detected. The abundances of panphytophygous microarthropods were not affected by treatments in either year.
To summarize, all treatments affected the abundance of at least one taxonomic group on at least one sampling occasion. Treatments, other than ammonium chloride and strepto-

mycin, affected at least one functional group. There was little consistency in effect, i.e. groups affected on the first date by a particular treatment were generally not affected on the second by that same treatment. However, the abundance of microarthropods was increased by ammonium sulphate and sulphuric acid on both occasions. In some instances diametrically opposed results were obtained. For example, the abundance of Astigmata was modified in nitric acid plots on both dates. On the first date their abundance was increased in the mineral soil (in comparison to the control plots) on the second, there was a decreased abundance in the organic layer (in comparison to sulphuric acid plots).

Individual species. The effects of the individual components of the "acid" rain on abundances of individual species are presented in Table 4. The abundances of twenty-nine species were altered by the treatments and all treatments had effects on more than one species. All effects discussed are significant at $p \leq .05$. Fourteen of the 29 species were affected by more than one treatment. The direction of the alteration in abundance of ten of these species was the same regardless of the treatment which affected the change. Also noted were four species, *Isotomiella minor* (Schäffer), *Stenacidia* sp. and *Pergamasus celticus* Bhattach. which were eliminated from plots sprayed with ammonium sulphate and two species, *I. minor* and *Phthiracarus membranifer* Parry eliminated from nitric acid treated plots after two years of spraying. *Isotomodes productus* (Axels.), *Geholaspsis aeneus* Krauss, *Brachychthonius jacoti* Evans, *B. immaculatus* Forsslund, *Liochthonius hystricinus* (Forsslund) and *Tydeus* sp. were present in one of the treatment plots but absent from control plots. Two species were found in both ammonium sulphate and urea plots which were not found in the controls. With the exceptions of *P. celticus* and *Megalothorax minima* (Will.), whose abundance increased in urea treated plots after one and two years of spraying, and *Liocthonius brevis* (Mich.), whose abundance increased in nitric acid plots on both sampling occasions the abundance of no other species was affected by the same treatment on both sampling occasions. The three most acidifying treatments, nitric acid, sulphuric acid and ammonium sulphate, affected the greatest number of species (see Table 4; nitric acid, 11 species; sulphuric acid, 12 species; ammonium sulphate, 11 species). Eight species were affected by more than one acid. The abundances of six of these species were affected in the same direction by the acids. However, of these 6 species a subset of 4 species were affected by other treatments and respond in the same manner.

Multivariate analysis

The ordinations derived from CCA of microphytophages from each soil horizon after one and two years are shown in Fig. 1 to Fig. 4. The results of Monte Carlo permutation tests (MCPT) performed on CCA data showed that overall differences emerged between the microphytophagous assemblages in the mineral soil after one year's spraying (first eigenvalue = .10, total inertia 1.79, $F = 4.09$, $p = .02$). Although a MCPT on the 1st axis of the CCA for soil microphytophages after two year's of spraying was not significant, a test of all axes was (eigenvalue of trace = .2, total inertia 1.6, $F = 1.31$, $p = .02$). Differences between these assemblages were found in the organic layer after the second year only (eigenvalue of 1st axis = .091, total inertia 1.7, $F = 4.16$, $p < .01$).

To separate the components of the assemblage reaction to the treatments, MCPTs were performed to compare the following: each treatment with the control, the two strong acid treatments (nitric and sulphuric) were compared, all nitrogen treatments were compared, all ammonium containing treatments were compared (Urea, which rapidly breaks down to yield ammonium, was included in this analysis), and finally, the two sulphur treatments were compared. The results of these analyses are given in Tables 5 and 6. When each treatment was compared with the control the significant responses found after one year's spraying were maintained in the second. Other differences emerged in the second year only.

Table 4. Effect of treatments on species after one and two years of spraying "components" of acid rain. "+" indicates that abundance was increased relative to the control plots; "−" indicates a reduction; "p" indicates that the species was present in the treatment but not in control plots and "e" indicates that the species was eliminated from treatments plots. Results are indicated at $p \leq .05$

	Nitric Acid		Ammonium Sulphate		Sulphuric Acid		Ammonium Nitrate		Urea		Ammonium Chloride		Streptomycin	
	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2	Year 1	Year 2
Collembola														
Frisea mirabilis	−										+		−	
Isotomiella minor		e		e				e				e		e
Isotomodes productus									p					
Lepidocyrtus lanuginosus				+						+		+		+
Megalothorax minima	−								+	+				
Stenacidia sp.				e										
Mesostigmata														
Geholaspis aeneus				p										
Olodiscus minima					+									
Pergamasus celticus				e		+			+	+				
Veigaia agilis											+			
Veigaia nemorensis		−				−				−		−		−
Cryptostigmata														
Brachychothonius jacoti												p		
Brachychothonius immaculatus			p											
Coccotydeus sp2	+		+								+			
Liochthonius brevis	+	+		+		+		+						
Liochthonius hystricinus										p				
Liochthonius simplex	−						−						−	
Oppia obseleta	+		+											
Oppia ornata	+		+		−									
Oppiella nova						+		+						
Oribatella meridionalis	+		+		+		+				+		+	
Phthiracarus membranifer		e												
Steganacarus magnus						+								

Prostigmata					
Alicorhagia sp		+			
Cocceupodes sp.		+		+	
Eupodes sp.		+			
Lorryia sp.		+			
Microtydeus sp. 2				+	
Tydeus sp.					p

Table 5. Results of CCA for microphytophages. The following tests, indicated by superscripts in row 1 of the table, were made using Monte Carlo Permutation tests; 1 treatment compared with control, 2 nitric acid contrasted with sulphuric acid, 3 all nitrogen containing treatment contrasted, 4 all ammonium releasing treatments contrasted, 5 ammonium sulphate contrasted with sulphuric acid

	Nitric Acid[1]	Amm. Sulphate[1]	Sulph Acid[1]	Amm. Nitrate[1]	Urea[1]	Amm. Chloride[1]	Strep[1]	Acids[2]	Nitrogen[3]	Amm.[4]	Sulph[5]
Microphytophages											
Organic layer year one											
Eigenvalue	.05	.033	.035	.043	.035	.044	.03	.043	.06	.048	.03
F value	1.9	1.39	1.47	1.83	1.46	1.95	1.30	1.80	2.45	1.63	1.09
P value	.16	.29	.24	.11	.25	.06	.37	.13	**.04***	.12	.66
Soil year one											
Eigenvalue	.059	.031	.06	.03	.03	.03	.03	.08	.08	.04	.06
F value	2.87	1.46	2.67	1.56	1.46	1.61	1.61	3.91	3.70	1.76	2.67
P value	**.01****	.28	**.02***	.09	.31	.21	.21	**.01****	**.01****	.38	.21
Organic layer year two											
Eigenvalue	.05	.04	.28	.03	.065	.04	.028	.05	.10	.10	.04
F value	1.91	1.69	1.14	1.26	2.76	2.31	1.17	1.90	4.11	4.07	1.61
P value	.12	.17	.69	.50	**.02***	.06	.58	.15	**.01****	**.01****	.30
Soil year two											
Eigenvalue	.051	.05	.054	.052	.051	.06	.055	.04	.05	.036	.04
F value	2.35	2.35	2.19	2.35	2.35	2.53	2.52	1.83	2.08	1.57	1.93
P value	**.01****	**.01****	**.02***	**.05***	**.05***	**.02***	**.02***	.19	.37	.58	.11

Table 6. Results of CCA for predators. Tests are as described in legend to Table 5

	Nitric Acid[1]	Amm. Sulphate[1]	Sulph Acid[1]	Amm. Nitrate[1]	Urea[1]	Amm. Chloride[1]	Strep[1]	Acids[2]	Nitrogen[3]	Amm.[4]	Sulph[5]
Predators											
Organic layer year one											
Eigenvalue	.08	.06	.09	.08	.09	.11	.09	.06	.11	.11	.07
F value	1.73	1.56	1.83	1.67	1.85	2.27	1.79	1.32	2.22	2.24	1.37
P value	.15	.32	.14	.18	.17	.08	.17	.34	.11	.09	.07
Soil year one											
Eigenvalue	.093	.09	.14	.08	.067	.07	.10	.12	.09	.05	.11
F value	2.94	2.98	4.41	2.60	2.08	2.08	3.14	3.68	2.84	1.51	3.67
P value	**.01****	**.01****	**.01****	**.02***	**.04***	**.05***	**.01****	**.01****	**.03***	.61	**.01****
Organic layer year two											
Eigenvalue	.047	.06	.05	.16	.03	.05	.08	.04	.16	.16	.05
F value	1.46	1.75	1.49	5.06	.90	1.70	2.65	1.11	5.12	5.01	1.40
P value	.4	.23	.30	**.01****	.86	.21	.06	.67	**.02***	**.02***	.32
Soil year two											
Eigenvalue	.05	.07	.06	.05	.04	.026	.07	.05	.08	.07	.05
F value	1.47	.93	.87	.69	.76	.67	1.81	1.48	1.98	1.82	1.46
P value	.38	.49	.64	.73	.85	.93	.21	.45	.40	.38	.38

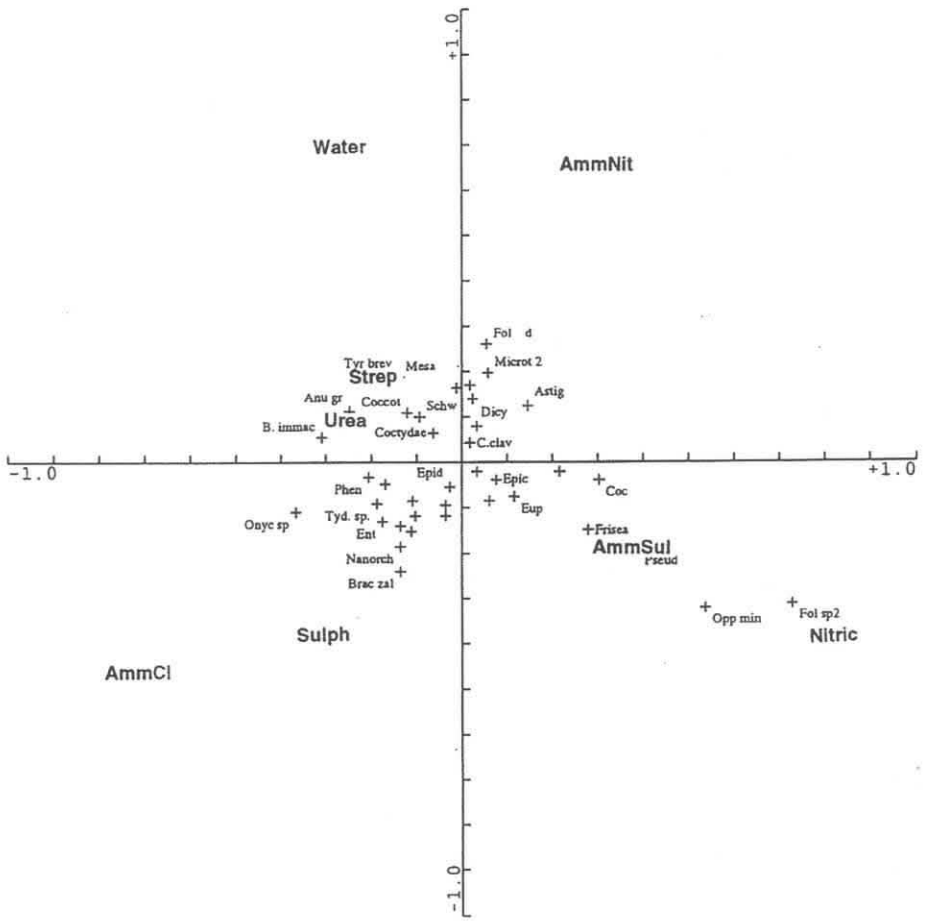

Fig. 1. CCA ordination of microphatyphages in litter after one year of spraying showing environmental variables (treatments) and species (species codes are given in Appendix 1)

In the organic layer, differences emerged between assemblages in nitric acid and the control plots, and between assemblages in sulphuric acid and the control plots after the second year. In the mineral soil differences between urea and control plots were found after both one and two years, while differences emerged between each treatment and control plots after two years.

After one year differences were detected between the microphytophagous assemblages inhabiting organic layer sprayed with nitrogen. When nitric acid was removed from the analysis (leaving the ammonium containing treatments) no differences remained. A similar result was found after the second year. Assemblages from plots receiving nitric acid also differed from those of the sulphuric acid plots after two years. A different picture emerges from the analysis of mineral soil inhabiting assemblages. Differences were detected, after one year between plots sprayed with nitrogen containing treatments. The differences were maintained even when nitric acid was removed from the analysis. After two years no differences in microphytophage assemblage composition were detected between nitrogen or ammonium treated plots.

There were no difference between the plots treated with sulphur compounds.

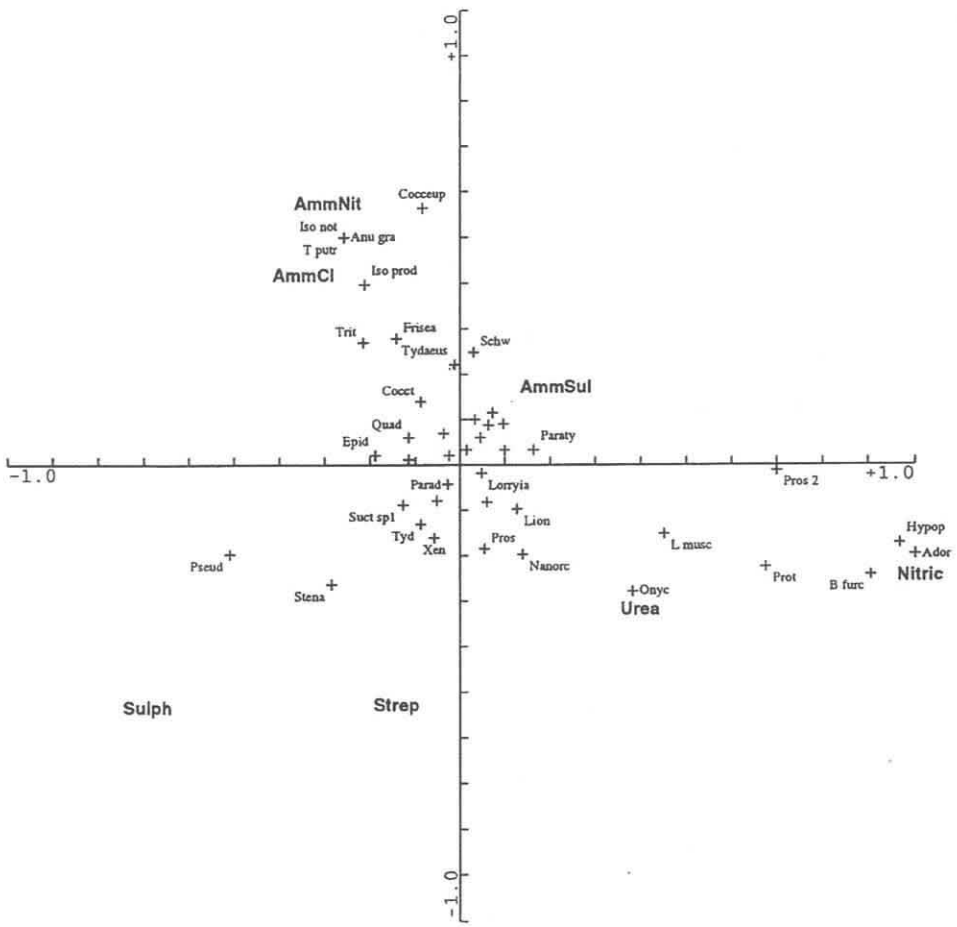

Fig. 2. CCA ordination of microphatyphages in litter after two years of spraying showing environmental variables (treatments) and species (species codes are given in Appendix 1)

Results from CCA of predators in the organic layer are similar to those of microphytophages. No overall differences were detected in the organic layer after one year but strong overall differences emerged after two years of spraying (1st eigenvalue = .16, total inertia 2.401, $F = 4.95$, $p < .01$). In the soil, overall differences in predator assemblages were detected after one year (1st eigenvalue = .172, total inertia 2.383, $F = 5.20$, $p < .01$) but no differences were found after two years.

The components of the response were analyzed using MCPTs, as described previously, and the results are shown in Table 6. No test revealed significant differences in predators in the organic layer after one year. After two years the predator assemblage composition differed in response to each treatment compared to the control. The assemblages from ammonium treated plots were similar to each other but the nitric acid assemblage was different (revealed by significant MCPT on all nitrogen-containing assemblages). This latter result conforms with that found for microphytophagous assemblages. For predators, as with microphytophages, the assemblages from the two strong acid treatments differed from one another after the second year.

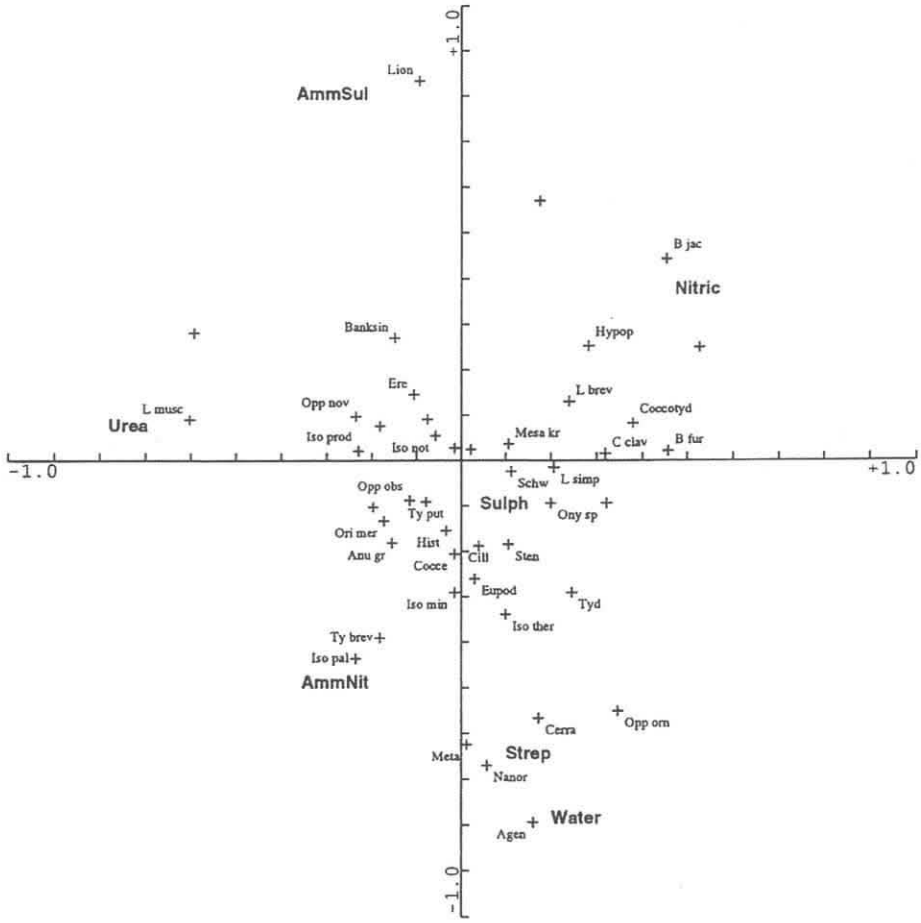

Fig. 3. CCA ordination of microphatyphages in soil after one year of spraying showing environmental variables (treatments) and species (species codes are given in Appendix 1)

Species richness and diversity

After one year of spraying no difference emerged in species richness or α diversity of microarthropods from the plots. After two years the α diversity in the organic layer of sulphuric acid plots was relatively high and that from nitric acid plots relatively low (Table 7). The differences in α diversity between these acidified plots was significant. The diversity in nitric acid treated plots differed from other plots sprayed with nitrogen compounds and differences between sulphur containing treatments were also found. Species richness was highest in urea plots and lowest in nitric acid plots. The difference in richness between nitric acid and other N treated plots was significant whereas no difference between these remaining plots was detected.

Table 7. Fisher's α diversity measure (means) and species richness (means) from organic and mineral soil after 1 and two years spraying with components of acid rain

Treatment	Fisher's α				Richness			
	Organic[1]	Min. soil[1]	Organic[2]	Min. soil[2]	Organic[1]	Min. soil[1]	Organic[2]	Min. soil[2]
N. Acid	9.47	6.21	5.72	4.18	27.89	21.7	20.66	13.88
Amm Sul	9.79	6.03	8.37	4.66	30.44	20.0	28.11	16.33
S Acid	6.29	6.36	11.10	5.01	30.88	18.00	24.33	17.33
Amm Nit	11.7	6.63	7.003	4.67	30.55	22.22	27.00	16.11
Urea	11.75	6.64	9.16	5.5	32.11	22.44	30.66	14.99
Control	11.01	7.97	7.49	4.63	28.33	19.44	22.77	13.66
Amm Chl	12.19	6.81	7.71	6.17	31.11	19.11	25.77	16.66
Strep	11.10	8.89	8.51	4.03	27.77	18.22	26.00	13.11

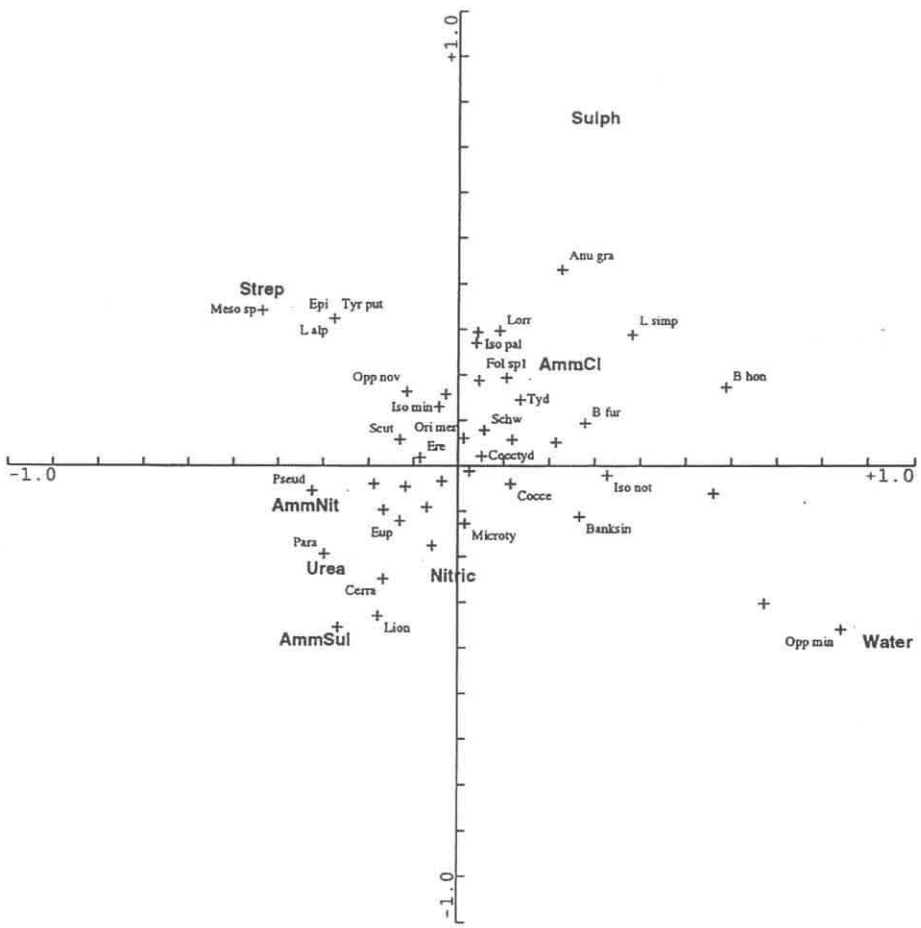

Fig. 4. CCA ordination of microphatyphages in soil after two years of spraying showing environmental variables (treatments) and species (species codes are given in Appendix 1)

Correlations between proton concentrations in organic layer and abundance of taxonomic groups, functional groups and individual species

The proton concentration of the organic layer, measured on the last day of the experiment (7 May 1991) was negatively correlated with abundances of Mesostigmata ($r = -.27$, $p < .02$). No other correlations with supra-specific taxonomic or functional groups were found.

The following species were positively correlated with proton concentration: *Amblyseius* sp. and *Microtydeus* sp. The following species were found to be negatively correlated with proton concentration: *Cilliba cassidea* (Herm.) ($r = -.36$, $p < .0004$), *Olodiscus minima* (Kramer) ($r = -.29$, $p < .02$), *Trachytes minima* (Trägårdh) ($r = -.27$, $p < .02$), *Veigaia planicola* (Berlese) ($r = -.38$, $p < .002$) and *Frisea mirabilis* Tullb. ($r = -.28$, $p < .0.2$). Correlations were found between the overall abundance of Collembola and Prostigmata ($r = .42$, $p < .001$; and between Astigmata abundance and that of Mesostigmata ($r = .30$, $p < .01$). Amongst functional groups a correlation was found between abundance of microphytophages and that of predators ($r = 0.32$, $p < .01$).

Bait Strips

Numbers of pierced lamina per bait strip for samples taken on 25 April 1991 and the associated test statistics are shown in Table 8 and 9. Differences were found in activity amongst the treatments. Strips in plots sprayed with nitric acid, ammonium sulphate and sulphuric acid (the most acidic treatments) differed from the control plots having reduced activity. No difference in activity was detected amongst these acidic treatments. Vertical activity amongst all treatments (Sy) was not found to differ (Table 10).

Discussion

Some comments on protocol

Even before its current use in acid rain research, simulating rainfall for environmental studies had become a common experimental procedure (Skiba et al. 1987). The two criticisms which are commonly levied at the manner in which simulated rainfall is applied in recent investigations, namely that the pH of the simulated rainfall is too low and that the amount of rainfall experimentally added is in excess of that which the plots receive naturally, (Skiba

Table 8. Number of pierced lamina per bait strip (Sx)

Treatment	Bait Lamina Strips															
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
N. Acid	3	2	2	5	2	1	2	0	3	0	1	3	2	3	1	4
Amm Sul	0	1	3	3	1	1	4	0	0	0	1	1	1	3	0	5
S Acid	2	2	3	1	1	3	0	4	3	1	0	2	2	0	0	2
Amm Nit	3	6	2	4	5	2	5	3	9	4	4	4	6	5	1	4
Urea	7	6	6	3	2	4	7	2	1	3	3	5	1	5	4	2
Control	6	1	4	2	4	3	8	4	3	4	7	5	5	4	2	4
Amm Chl	2	5	0	4	1	3	3	4	3	2	4	5	3	6	2	0
Strep	4	6	4	3	1	3	4	1	3	5	2	4	5	3	5	0

Kruskal-Wallis Test for treatments
$\chi^2 = 36.23$ $p < .001$

Table 9. Differences in Sx (number of pierced lamina per strip) between treatments tested by Kruskal-Wallis non-parametric test

	χ^2	prob $>\chi^2$	Level of significance
Nitric Acid [1]	9.49	.002	**
Ammonium Sulph [1]	12.65	.000	***
Sulphuric Acid [1]	13.609	.000	***
Ammonium Nitrate [1]	0.18	.89	n.s.
Urea	24	.62	n.s.
Ammonium Chlor [1]	2.84	.091	n.s.
Streptomycin [1]	1.12	.29	n.s.
Acids [2]	2.11	.35	n.s.
All Nitrogen [3]	27.205	.000	***
All ammonium [4]	20.79	.000	***

[1] Sx of treatment compared with Sx of control.
[2] Sx of acidifying most treatments (Nitric Acid Sulphuric Acid, and Ammonium Sulphate) compared.
[3] Sx of all nitrogen containing treatments compared.
[4] Sx of all NH_4 containing treatments compared

Table 10. Number of pierced lamina per position on bait strip (Sy)

Treatment	Bait Lamina Strips															
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
N. Acid	12	5	4	2	3	3	0	1	0	0	0	0	0	2	2	0
Amm Sul	7	4	6	2	2	1	1	0	1	0	0	0	0	0	0	7
S Aci d	4	3	4	5	2	3	2	1	1	1	0	0	0	0	0	0
Amm Nit	15	15	12	9	7	3	2	1	1	0	0	0	0	2	0	0
Urea	13	13	8	4	4	6	1	3	4	1	0	1	3	0	0	0
Control	10	14	13	8	4	4	6	1	3	4	1	0	1	3	0	0
Amm Chl	11	11	11	6	2	3	0	0	1	1	1	0	0	0	0	0
Strep	13	12	10	5	6	4	1	0	1	1	0	0	0	0	0	0

Kruskal-Wallis Test for treatments
$\chi^2 = 4.40$ p $<.73$

et al. 1987), could apply to the experiment reported here. However, the protocol followed in the current experiment was designed with the following in mind: the levels of N and S input were to be identical (150 kg N $ha^{-1}a^1$ and 171.4 kg S $ha^{-1}a^{-1}$) in the appropriate treatments to permit direct comparisons between the treatments and with results reported in the literature. The amounts of N and S, though greater than those received even in areas subject to extreme pollution, must be considered low compared with some previous studies. The treatments were applied in a fine mist to simulate input from rain. This contrasts with the single large applications of N fertilizers often associated with silvicultural research, the results of which are useful for the purposes of comparison. These silivicultural experiments, if classified according to the terminology associated with stress ecology (see Bender et al. 1984; Calow & Berry 1989), could be considered as "pulse" perturbations, i.e. the perturbation is applied in a single input and responses are followed through time. The present experiment was designed to evaluate the impact of the components of a polluted rain upon the abundance and diversity of species and the design thus attempted to approximate more closely the "press" type perturbation (where the perturbation was unrelenting over the course of an experimental period), deemed to be more realistic in the context of atmospheric-borne pollution.

The conjecture was formulated and tested in this study that polluted rain, which has a regionally diverse chemical composition, could affect microarthropod assemblages in a manner which related not only to pH, but which related also to the chemical species in the rain.

Treatment effects

The influence of stressors on biological systems has, for reasons that are obvious, been the subject of a vast number of studies. Generalized models predicting the responses of individual organisms to stress have been presented (Selye 1973). An application of a "pulse" typically leads to a measurable negative response, followed by a positive compensation and a final return to normal activity (Gray 1989). These responses are typically physiological ones. Responses at the population level are less easy to predict and discussion is made complicated by the multiple meanings of the term "perturbation" (Underwood 1989) and the ambiguities associated with the term "stress" (Grime 1989). This semantic concern notwithstanding, it is clear that "press" experiments are likely to cause quite different patterns and time courses in the response of species (Bender et al. 1984). It is often difficult, therefore, to determine, for "press" experiments, when the experiment should be terminated because stressed populations of animals often oscillate and can thus converge with controls, although the convergence may be ephemeral (Underwood 1989).

For the present purposes it is worth considering that the assemblage of microarthropods at the site chosen for this experiment developed in response to a "natural" experiment. That is, the sitka spruce was planted as a monoculture on land which was previously arable. Although some work is currently being carried out on the origins of some aspects of the fauna in response to the planting of conifers in Ireland (e.g. carabids, Coll et al. (1995)), no comparable surveys of microarthropods exists for Irish material. The nutrient amendment, added as part of the factorial experiment, may serve to relieve stress on already stressed biota. Since the planting of conifers on land with diverse histories of usage is now quite commonplace, the results of the present investigation may be of general interest.

The results demonstrated that changes in both faunal abundance and diversity were associated with the different treatments. Effects of treatments on the abundance of 14 species were detected after one year whereas 18 species were affected after year two. All treatments affected the abundance of three or more species. There was however, little consistency of effect from one year to the next. Nitric acid treatment lead to an increase in *Liochthonius brevis* abundance after both year one and two. Urea increased the abundance of *Pergamasus celticus* and *Megalothorax minimus* after both years.

The negative response of *Megalothorax minimus* to nitric acid (in the mineral layer after one year) and *Veigaia nemorensis* (Koch) to both nitric acid and sulphuric acid (in organic layer and mineral soil after two years) is similar in direction to that found by Hågvar & Kjondal (1981) in response to the application of sulphuric acid. The negative reaction of *Isotomiella minor* (which disappears from nitric acid treated organic layer after two years) and the positive reaction of the *Steganacarus magnus* (Nich.) (in the organic layer of sulphuric acid plots after two years) and *Pergamasus celticus* (in the organic layer after two years) populations resonate with observations made by Hågvar (1984), Hågvar & Kjøndal (1981). The positive responses of *Oppia ornata* (Oudemans) and *O. obseleta* (Paoli), however, contrast with those found by Bååth et al. (1980).

The general lack of species common to all studies makes explicit comparisons with fertilizer experiments, in that respect, difficult. The negative reaction of *Isotomiella minor* to nitric acid, ammonium sulphate, ammonium nitrate and ammonium chloride after two years resonates with the negative reactions of this species to applications of ammonium nitrate found by Lohm et al. (1977) two and a half years after application. The fact that the species responded to the treatments regardless of the different pH associated with the applications suggests that the ammonium component is critical. It can be seen (Table 11) that most of the effects were enhancement in abundance.

Effects on the abundance of Collembola, Cryptostigmata and Astigmata were noted. All treatments had an effect of either raising (sulphuric acid, ammonium sulphate, ammonium nitrate, nitric acid and urea) or lowering (nitric acid and streptomycin) of faunal abundance in at least one soil layer in either year one or year two (or the two combined). There was little consistency of effect, however, over the two years. Effects on abundance observed in one soil portion after one year were not maintained after two years. Effects of some treatments emerged on the second sampling date only.

The variable response of total Collembola to acid treatments (lowered in nitric acid plots, increased in sulphuric acid) is interesting considering that Hågvar (Hågvar 1984; Hågvar & Kjøndal 1981) found a positive response to sulphuric acid in a field experiment. The finding here, that Collembola populations respond in opposing ways to the acid treatments, suggests that the chemical composition of the acid (the treatments had similar pH) may be critical.

The response of organic layer dwelling Cryptostigmata to ammonium sulphate (after one year) and ammonium nitrate (after one and two years) requires consideration. Strong acidification was found by Hågvar & Kjøndal (1981) to have increased the abundance of Cryptostigmata. Although ammonium sulphate is acidifying, ammonium nitrate does not lower pH much below that of control plots. Therefore, the positive response found in the present experiment possibly reflects the effects of nutrient enrichment, in the form of ammonium, from both the ammonium sulphate and ammonium nitrate, as much as it does the effects of acidity. Behan et al. (1978) resported an initial "shock effect" in microarthropod response to urea additions followed by an increase in abundance. This was in conformity with the expectation for species effect as a consequence of a "pulse" type experiment.

Increased abundance of Astigmata in the mineral soil of plots receiving nitric acid for one year was similar in direction to responses of this group to acidification with sulphuric acid over a 3 month period reported by Hågvar & Kjøndal (1981). However, a longer term pattern of reduction in abundance emerges after two years when nitric acid and sulphuric acid plots tended to have a lower abundance in the organic layer, when compared with controls. When the abundance from both sets of plots was considered simultaneously, a reduction, relative to all other plots, was detected (see Table 11). The increased abundance in ammonium sulphate, urea and ammonium chloride plots compared to the controls corresponds with a similar increase reported by Behan et al. (1978) in response to urea fertilization. They considered this increase to reflect a rapid increase in food supply.

When treatments affected the abundance of Prostigmata, relative to the control, these were enhancements except in the case of streptomycin plots where abundance in the mineral layer was halved. In contrast a negative effect of sulphuric acid was found by Hågvar & Amundsen (1981). Huhta et al. (1986) also report a negative effect of urea on Prostigmata abundance.

The results of the CCA indicated that the spraying of the different component of "acid" rain, or the different nutritive compounds on the soil lead, in some cases, to the development of assemblages which were distinct from assemblages developed in the absence of exogenous inputs. In the case of microphytophages, assemblages developed in the organic layer of plots sprayed with nitric acid and sulphuric acid which were distinct from those in the control plots. In addition, these assemblages were distinct from one another. The difference in these assemblages was apparently confirmed by the diversity difference detected in the nitric and sulphuric acid plots (each differed from the control and also differed from one another). In the soil at the end of the second year of spraying, assemblages developed in the plots of all treatments which were different from the assemblages in the control plots. In this case, however, the assemblages did not differ among each other (i.e. using CCA no differences were detected amongst the assemblages in plots receiving acids, nitrogen compounds, ammonium releasing compounds or sulphur compounds when these groupings were examined). The predator assemblages which developed after two years confirm this impression to some degree. In the organic layer the predator assemblage in each treatment was distinct from the assemblage in control plots. In this case, the two acid treatments

Table 11. Summary of treatment effects on abundance and community attributes (results from Monte Carlo Tests and diversity measures). Except where otherwise indicated comparisons are of treatment and control. (O = organic layer; M = mineral soil; Y1 and Y2 are samples taken one and two years after initiation of experiment respectively; ↑ = abundance increase; ↓ = abundance decrease; △ = changes in community composition compared to control; α = Fisher's α diversity; R = species richness). Emboldened symbols indicate comparison with control plots. When other comparisons are made the result is indicated by reduced font

	NA	AS	SA	AN	U	AC	Strep
Abundance							
Collembola[1]	↑ OY2		OY2 ↓				
Crypto		↑ OY1		↑ OY2			
Astig[2]	O+MY2 ↑ ↓ MY1	↑ OY2	O+MY2 ↓		↑ OY2	↑ OY2 O+MY2	
Prostigs	↑ ↑ MY1 MY1	↑ MY2	MY1 ↑ ↓ MY2 M+OY2	↑ OY2 M+OY2	↑ OY2 M+OY2		MY1 ↓
Mesostigs							
Microphy[3]	↑ MY1	↑ OY1 O+MY2	↑ OY1+Y2	↑ OY2 O+MY2	↑ MY1		
Macro[4]	OY2 ↓						
Pan							
Preds		↑ MY2					
CCA							
Microphy[5]	△ OY2 MY2	△ MY2	△ MY2 OY2	△ MY2	△ MY1 MY2	△ MY2	△ MY2
Preds[6]	△ OY2	△ OY2	△ OY2	△ OY2	△ OY2	△ OY2	△ OY2
Diversity							
Overall	R+α OY2 ↓		↑ α OY2		↑ R OY2		

[1] Collembola from organic layer of NA and SA plots differ from one another but neither differ from the control.
[2] Differences in abundance between plots sprayed with acids (SA and NA) and other treatments significantly lowered in organic layer after two years.
[3] Differences in abundance amongst ammonium containing treatments when organic and mineral layers are combined.
[4] Difference between macrophytophage abundance in NA and SA plots observed but difference between the effect of these acids and all other treatments also detected.
[5] NA differs from SA in organic layer after both one and two years. Differences detected in assemblages after one year in mineral soil when N containing and ammonium containing treatments are compared. These differences have disappeared by year two.
[6] NA assemblage differs from SA assemblage and NA differs from all other N treatments after two years.

lead to divergent assemblages as did the two sulphur treatments. Differences between the assemblages in ammonium treated plots were not detected.
It is apparent that low level chronic input of nutrients to the soil will affect changes in the assemblages of resident microarthropods. The response of assemblages to ammonium compounds is often similar. On occasions when differences between assemblages responding to applications of ammonium releasing compounds were detected, the assemblage developed on only one of these ammonium treated plots were different from the control (i.e. urea for microphytophages in the organic layer after two years, and ammonium nitrate for predator assemblages in the soil after one year). Further confirmation that the ammonium treatments lead to similar assemblage development, comes from an observation that on three of the five times when differences were detected amongst nitrogen treatments the differences disappeared when nitric acid was removed from the analysis (see Tables 9 and 10). Assemblages of both microphytophages and predators developed in the organic layer of nitric acid plots were different from those developed on sulphuric acid plots after to years and both sets of assemblages differed from the controls.
Craft & Webb (1984) in their examination of the effects of acid and non-acid sulphate salt solutions on forest floor arthropods found some highly variable responses to their treatments, some of which they attribute to the fertilizing effects of K^+ on microorganisms.
It is worth pointing out that none of the treatments applied seemed to have strongly toxic effects on the fauna as a whole, i.e. there was no evidence of any one treatment strongly and consistently depleting faunal groups. In this light it can be noted that streptomycin, considered to be bactericidal, affected the abundance of *Frisea mirabilis*, which, with its piercing mouthparts may be a bacterial feeder.

Ultimate causes of changed abundance

The elucidation of the ultimate causes of the changes in the abundance of several groups was not a principal objective of this experiment. It is now widely accepted that pH, which can have important effects on microarthropod reproduction (Hutson 1978), is not the exclusive determinant of changes (Huhta et al. 1986). Hågvar (1990) has shown that pH alters the competitive ability of acidophilic species allowing them to dominate at lower pHs. The great variability of responses amongst the microflora (Fritze 1992) to environmental pollution suggests that if competitive ability is important in maintaining the abundance of individual species and consequently in determining microarthropod assemblage structure, then these environmental changes which affect the trophic base of the assemblages (and, as we have seen, for the assemblage discussed here 80% + are microphytophages) will have an unpredictable effects. Furthermore, Yodzis (1988) has shown that indirect effects ("where species A affects species C through a chain of intermediate species" Yodzis 1988) when modeled show that "press" perturbations have highly indeterminate outcomes.
It has become quite common practice, when establishing the collective responses of the fauna, to group species into supra-specific taxonomic categories, which, as we have seen above, is a useful comparative approach. However, in an attempt to see the extend to which effects of a pollutant are resource-determined (populations controlled by "bottom-up" forces) it is more useful to group species according to their trophic behaviour. When species were grouped in this way for the present experiment, significant positive responses of microphytophage abundance were found in both years. Increases in the abundance of predatory microarthropods in the mineral portion of ammonium sulphate treated plots corresponds to the increases in the abundance there of microphytophages, which presumably are their prey. The overall correlation between predator numbers and microphytophages is the only correlation between supraspecific groupings found. These results are suggestive of regulation through overall increases in microbial biomass.
The decrease in microphytophage abundance detected in response to nitric acid inputs may reflect a decrease in the palatibility of the organic layer.

Correlation analysis showed that Mesostigmata abundance related negatively to pH. This overall relationship emerged from the collective contributions of *Cilliba cassidea*, *Olodiscus minima*, *Trachytes minima* and *Veigaia nemorensis* all of which were also negatively related to pH.

It would be impossible to predict the long term outcome of a continuation of such chronic nutrient and acid additions to the microarthropod assemblages in the forest soil. Some studies have charted the progress of populations as they recover from a single application of stress (i.e. "pulse" type perturbations). There is no study, however, that we know of, which monitors microarthropod population responses to a chronic stress (i.e. "press" type perturbations) for longer than 6 years. Due to a possibilty of a large numbers of indirect interactions in detritus-based assemblages it is worth bearing in mind Yodzis's (1988) conclusion that "pulse" perturbations conducted over the short-term are inconclusive when long-term predictions are required.

It should be borne in mind that microarthropod feeding can be sporadic and that analysis of gut contents reveals that up to 89% of Collembola, at least, have empty guts at any one time (Usher 1985). This may represent a "nonfeeding phase of the intermoult period" (Muraleedharan & Prabhoo 1978, cited in Usher 1985). If the effects of polluted rain are mediated through soil solution chemistry and the microbial population, then it is possible that this "resting period" will delay any of the expected consequences for microarthropod populations. In the light of this, the reduction of feeding activity, measured by bait strips, in plots which received acid treatments may be indicative of a potential for more radical population changes in the long-term.

This study confirms the suspected multiplicity of effects attributable to "acid" rain. Although the application of strong acids provoked the strongest response from the biota (both in terms of the number of species effected and in the observed responses of the assemblage) it was found that the spraying of all components of acid rain had biotic consequences. The impact of the sorts of modest changes to the structure of the microarthropod community which we have demonstrated here for the regulation of important ecosystem processes are discussed in Heneghan & Bolger (in prep). We have shown that processes occurring in the soil at spatial and temporal scales similar to the ones in which microarthropods participate (i.e. the fluxing of labile nutrients in the organic layer) can be affected by changes in the assemblages imposed by the environmental perturbation.

Appendix 1:. Abbreviations of species appearing in ordinations

Species	Abbreviation in ordinations
Mesostigmata	
Cilliba cassidea (Herm.)	Cill
Prostigmata	Pros[1]
Alicorhagia sp.	Alic
Cocceupodes clavifrons (R. Can.)	C clav
Cocceupodes sp.	Cocceup
Coccotydeus sp. 1	Cocctydae
Coccotydeus sp. 2	Cocct
Ereynetes sp.	Ere
Eupodes sp.	Eup(od)
Lionopodes sp.	Lion
Lorryia sp.	Lor
Nanorchestes arboriger (Ouds.)	Nanorc
Paratydeus sp.	Paraty
Protereunetes sp.	Pro/t
Rhagidia sp.	Rhag

Appendix 1:. (Continued)

Species	Abbreviation in ordinations
Scutacarus sp.	Scut
Triophtydeus sp.	Trit
Tydeus sp.	Tydeus/tyd
Cryptostigmata	
Adoristes poppei Koch	Ador
Banksinoma lanceolata (Mich.)	Banksin
Brachychthonius furcata Weis-Fogh	B fur
Brachychthonius immaculatus Forsslund	B immac
Brachychthonius jacoti Evans	B jac
Brachychthonius zelawaiensis (Sell.)	Bra zal
Ceratoppia bipilis (Herm.)	Cerra
Epicriopsis sp.	Epic
Epidameus sp.	Epid/Epi
Liochthonius brevis (Mich.)	L brev
Liochthonius muscorum (Forsslund)	L musc
Liochthonius simplex (Forsslund)	L simp
Oppiella nova (Oudemans)	Opp nov
Oppia minus Paoli	Opp min
Oppia obsoleta (Paoli)	Opp obse
Oppia ornata (Oudemans)	Opp orn
Oribatella meridionalis Berlese	Ori mer
Paradameus clavipes (Herm.)	P clav
Quadroppia sp.	Quad
Suctobelbella sp.	Suct sp1
Collembola	
Agenia sp.	Agen
Anurida ellipsoides Stach.	An ell
Entomobrya sp.	Ent
Folsomia sp.	Fol sp1
Folsomia sp. 2	Fol sp2
Frisea mirabilis Tullb.	Frisea
Isotoma notabilis Schäffer	Iso not
Isotomina thermophila Axels	Iso ther
Isotomodes productus (Axels.)	Iso prod
Isotomurus palustris (Müller)	Iso pal
Megalothorax minimus (Will.)	Megaloth
Mesaphorura krausbaueri Born.	Mesa kr
Metaphorura sp.	Meta
Micranurida	Micr
Onychiurus sp.	Onyc sp
Phenotrix sp.	Phen
Pseudanurophorus sp.	Pseud
Sphaeridia sp.	Sphaer
Stenacidia sp.	Stena
Tomocerus minor (Lubb.)	Tom min
Tullbergiinae	Tull
Xenyella humicola (Fabr.)	Xen
Astigmata	Astig[1]
Schwiebia sp.	Schw
Histiostoma sp.	Hist
Tyrophagus putrescentiae (Schrank)	T putr
Hypopus	Hypop

[1] Code for unidentified species of Prostigmata and Astigmata

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